REMARKS

Claims 1, 4-12, 14, 15, 28, 30-36 and 43 are pending in the application. Claims 1, 2, 4, 5, 6, 7, 9, 10, 11, 14, and 28 have been amended. Claims 3, 13, and 29 have been cancelled.

The amendments to the specification include a renumbering of the tables, which presents the tables in the numerical order that the tables are presented. These amendments are made to overcome the objection to the specification on page 6 of the current office action because the tables were not numbered in the order presented. This renumbering of tables does not constitute new matter. Additionally, Table 1 on page 6 of the specification is amended to show that SEQ ID NOS. 4 and 5 commence with valine at position 869, not 859 as formerly recited. This amendment is made to correct a typographical error and does not constitute new matter, as is apparent from the identity of SEQ ID NO. 1 beginning at position 869 with SEQ ID NOS. 4 and 5, respectively, at position 1. Page 17 at line 13 has been amended to correct the citation to Tomme and Claeyssens by deleting an extraneous "l."

Claim 1 has been amended to recite the contents of Table 5 on page 35 of the amended specification. This amendment is made because the examiner has objected to the specification as not teaching consensus sequences for the homology claims. Claim 1 now recites also recites 70% homology to SEQ ID NO. 3, for example, as supported on page 19 at lines 3-4 of the amended specification. Each of claims 1, 2, 3, 4, 5, 12, and 14 have been broadened by deleting the word "thermostable," as the4 examiner appears not to give this word any patentable weight. Claims 4, 5, 6, 7 and 8 have been amended to correct the dependencies to claim 1, as claim 3 has been cancelled. As claim 1 now

recited 70% homology, claim 6 has been amended to clarify identity with SEQ ID NO. 3. claims 10 and 11 have been amended to recite, respectively, 90% and 80% identity with respect to SEQ ID NO. 3. Claim 28 has been amended to recite a Markush Group that is related as segmented components of SEQ ID NO. 1, and to delete reference to percent identity.

The objection to the Specification on page 2 of the Office Action dated March 11, 2003 is overcome by the amendments to claims 1 and 28. It is alleged that the specification does not teach the consensus sequences to identify sequence identity percentages with respect to AviIII. Claim 1 has been amended to specifically recite the positional alignments of sequences between GH74_Ace and AviIII_Aac shown in the Table 3 on page 34 of the application as filed. This alignment comparison was produced using the ClustalW program, as reported on page 33 of the application as filed, beginning at line 9. Similar comparisons may, for example, be obtained using this program or similar programs to identify all of the GH74 family consensus sequences. It is sufficient to overcome the objection that claim 1 has been amended to recite the alignments between GH74_Ace and AviIII_Aac, and claim 28 has been amended to delete reference to percent identity. The amendment to claim 1 recites the motif or conserved sequences by which the GGH74 domain can be identified and, accordingly, overcomes the objection to the specification.

Claims 1-9, 14, 15, 28-36 and 43 stand rejected under 35 U.S.C. §112, first paragraph, for lack of enablement. It is alleged that these claims, including at least claims 1, 14 and 15, are not supported by a specification that teaches any thermostable AviIII peptide having a GH74 domain. As to claims 1, 14, and 15, the Examiner asserts

that knowledge and guidance are required to determine which amino acids, if any, are tolerant of modification, as well as those which are conserved and presumably not subject to modification. This issue has been resolved by deleting the word "thermostable" from claims 1, 2, 4, 5, 12, 13 and 14 in each instance, and by amending claim 1 to recite the positional identity of sequences in comparison to AviIII_Aac shown in Table 3 on page 34 of the application as filed.

The amendments to claim 1 are made to show that Applicants did disclose which amino acids are tolerant of modification. Applicants previously traversed the rejection because such information is provided, for example, on pages 32-33 of the Specification in Example 2 and Table 3, which provide those skilled in the art with sufficient information to make these determinations by observing the conserved sequences among the GH74 family. Now that comparison is specifically claimed in claim 1. The remaining objections as to claims 2-5, 6-9, 28-36, and 43 are postulated for identical reasons, and Applicants similarly traverse these objections. The recitation in claim 1 of 70% sequence identity is made in context of preserving these conserved sequences.

Page 4 of the Office Action dated March 11, 2002 further rejects claims 1-9, 14 and 15, as well as claims 28-36 and 43 under 35 U.S.C. §112 first paragraph for the reasons explained in the office action dated August 1, 2002. As to claims 1-9, 14, and 15, the substance of the rejection is that only a single representative species of the claimed genus is disclosed in the written description, yet the genus pertains to any GH74 catalytic domain peptide having a CBDIII domain.. Amended claim 1 overcomes the rejection by specifically reciting which sequences are tolerant to modification. Examples 2 and 3 on pages 33-35 of the specification as filed show a rationale for observing the conserved

identities between GH74_Ace and AviIII_Aac, as is now specifically recited in claim 1. Example 3 mentions that fusion proteins and site-directed mutagenesis may be availed, for example, to provide a variety of sequences in a genus context. Applicants have disclosed and claimed a rationale for making the claimed genus by virtue of the conserved sequences recited in claim 1, and the segmented combinations of claim 28. As such, Applicants have disclosed more than a single working embodiment.

Whether such changes may be made according to the 70% homology of claim 1 while preserving thermostability is now a moot issue, since thermostability has now been deleted from the claims in each instance. Even so, thermostability is an inherent characteristic of claims 6, 9, 12, and 28, which call out identity with respect to sequences that are known to be thermostable.

Claims 28-36 and 43 are further rejected because the claims are directed to a genus of sequence homology, e.g., 70% identity with SEQ ID No. 1, but none of the homologous sequences are specifically disclosed. Claim 28 has been amended to delete reference to this homology and, consequently, overcomes the rejection. Claim 43 depends from claim 1 where the objection is overcome by the different recitation, as discussed above.

Claims 1-13, 28, and 29 stand rejected under 35 U.S.C. §103(a), as being unpatentable over Mohaghegi et al. 1986 in view of 'Berghem et al. 1976 and Katz et al. 1968. Mohaghegi et al. 1986 is said to show the isolation of *Acidothermus cellulolyticus*, but not the isolation of cellulase therefrom. Berghem et al. 1976 is used to show the isolation of endoglucanase from *Trichoderma viride*. Katz et al. supposedly shows motivation to combine, since it is desirable to generate alternative cellulases capable of

commercial scale processing at elevated temperatures. Although Bronnenmeir et al. 1991 is not cited as a reference in formulating the §103 rejection, the Examiner applies Bronnenmeir et al. 1991 to show that exoglucanases can degrade Avicel, and states that "[A]ffinity chromatography on an Avicel column would be a very powerful method for isolating the cellulase of the instant application, as it was well known in the art that exoglucanases bind to cellulose resin." To this end, the examiner also applies Tan et al., 1986, which is also not included in the combination of references cited in support of the §103 rejection.

It is not sufficient that the Examiner can propose a modification of Berghem et al., via Bronnenmeir et al and Tan et al., to show other methods that have been used to isolate exoglucanases and endoglucanases as a class of materials. The examiner must specifically show isolation of a GH74 family exoglucanase having the sequences that are specifically claimed. "To establish prima facie obviousness of a claimed invention, all the claim limitations must be taught or suggested by the prior art." *See* MPEP 2143.03. At present, no reference teaches or suggests the sequences that are specifically claimed, for example, that of SEQ ID NO. 3 (claims 1, 6, 9, 10 and 11), or SEQ ID NO. 4 (claims 7 and 9), SEQ ID NO. 5 (claim 8). Even in combination, the references do not show any of the sequences that are specifically recited in amended claim 28.

As to claim 12, none of the references teach or suggest the sequence of SEQ ID NO. 1, which is inherently *thermostable*. Furthermore, none of the references, even if Bronnenmeir et al and Tan et al. are included, teach a GH74 family exoglucanase. It could not have been predicted at the time of the invention that those skilled in the art would successfully locate a GH74 family exoglucanase from *A. cellulolyticus*, much less

the sequence that is specifically recited in claim 12. Furthermore, these same arguments attach to identity with the GH74 domain, as recited in claim 6; the full length sequence of claim 12, and the CBDIII domain recited in claims 7 and 8.

The present combination of Mohaghegi et al. 1986 in view of Berghem et al. 1976 and Katz et al. 1968 uses Berghem et al. to show the isolation of a cellulase, but it is an endoglucanase. As Paragraph 9 of the Rule 132 Declaration filed December 26, 2002 makes clear, the claimed GH74 domain functions as an exoglucanase, not an endoglucanase. Therefore, the combination does not teach or suggest all of the claim limitations because the combination, if proper, would merely result in the isolation of an endoglucanase from *A. celllulolyticus*. This does not teach the isolation of an exoglucanase. Furthermore, the combination of references does not teach the isolation of an exoglucanase having the specific sequences recited in the claims, as noted above

The applicability of Bronnenmeir et al. and/or Tan et al. is unclear in context of the rejection. In essence, the Examiner asserts that the GH74 sequences which are specifically called out, for example, in claims 1, 6 , 9, and 12 are made obvious by the "powerful method" of columnar chromatography with Avicel since both endoglucanases (Berghem et al., Irwin et al.) and exoglucanases (Bronnenmeir et al., Tan et al.) can degrade Avicel. Supposedly, this is meant to show that "Berghem et al. teach an assay that would have utility in analyzing the activity of the cellulase of the instant application during biochemical purification." We respectfully disagree.

First, at the time of the invention, no one could have predicted that Applicants would discover a new GH74 family enzyme, much less one having the recited sequences. If the examiner means to modify Berghem et al. with Bronnenmeir et al. and/or Tan et al.,

then he must specifically do so, observing all of the formalities of why the art suggests that the modification be made to show the claimed sequences.

Berghem et al. reported isolating an endoglucanase, not an exoglucanase. Berghem et al. is actually nonanalogous art. that is not properly combinable to show isolation of an exoglucanase. This is because those skilled in the art would not attempt to follow Bergheim et al. when attempting to isolate an exoglucanase, when Berghem et al. merely taught the isolation of an endoglucanase. Even assuming that Bergheim et al. can be properly combined, there is nothing specific in any reference of record that shows the specific sequences recited in the claims, as noted above.

The examiner alleges that the techniques taught by Berghem et al. will also work to isolate exoglucanases. It is not the case that those skilled in the art attempting to isolate an endoglucanase will serendipitously discover an exoglucanase using the same means. Other researchers, notably Tucker 1992 and Adney 1994 engaged in *Acidothermus cellulolyticus* research where they merely discovered endoglucanases. Besides, where endoglucanases and exoglucanases both have affinity for Avicel, this would not serve to isolate either type of enzyme exclusive of the other.

GH74 family enzymes are not highly conserved in the catalytic domain. The attached Rule 132 Declaration from Dr. Himmel documents computer research showing that, among 12 reported GFH74 family sequences, the highest rate of homology is about 50%. Claim 1 distinguishes the art by reciting at least 70% sequence identity. Claims 12 and 28 recite specific sequences. The remainder of the claims depend from claims 1, or 28 and are likewise allowable, also having patentable merit of their own as noted above.

Applicants' attorney respectfully solicits a Notice of Allowance in this application. The Commissioner is authorized to charge any additionally required fees to deposit account 14-0460. Should the Examiner have any questions, comments, or suggestions that would expedite the prosecution of the present case to allowance, Applicants' representative, Paul White, earnestly requests a telephone call at (303) 384-7575.

Respectfully submitted

Paul J. White

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Dated: June 17, 2003.

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#19

PATENT
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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant(s)	Shi-You Ding et al.		
Serial No.:	09/917,376	Group No.:	1652
Filed:	July 28, 2001	Examiner:	Swope, Sheridan
For:	Thermal Tolerant Avicellase From *Acidothermus cellulolyticus*	Confirmation No.	9956

CERTIFICATE OF MAILING

I hereby certify that this paper (along with any paper referred to as being attached or enclosed) is being deposited in the United States Postal Service on the date shown below with sufficient postage for first class mail, in an envelope addressed Commissioner for Patents, U.S. Patent & Trademark Office, P.O. Box 1450, Alexandria, VA 22313-1450.

June 17, 2003
Date

Brenda E. Brantley
Brenda E. Brantley

Commissioner For Patents
P.O. Box 1450
Alexandria, VA 232313-1450

RULE 132 DECLARATION OF DR. MICHAEL E. HIMMEL

This Declaration is presented to provide the Examiner with additional evidence that deserves consideration in traversing one or more of the rejections presented in an Office Action dated March 11, 2003.

1. I am the Michael Edward Himmel, PhD, who is also a named inventor in the above-identified patent application.

2. I am employed as a scientist at the National Renewable Energy Laboratory located in Golden, Colorado where I hold the position of Principal Scientist. I am engaged in research that includes the identification, isolation and cloning of cellulases.

3. I have reviewed the claims, as amended, in addition to the office action dated March 11, 2003.

4. Exhibit A to this declaration is a computer printout of comparison data that I produced using the BLAST program to compute identities between the catalytic domains of reported GH74 family nucleic acid sequences. The basis of comparison was the GH74 catalytic domain that is reported as SEQ ID NO 3 of the present application. I used the BLAST program to compare identities between this domain and ten GH74 catalytic domain sequences that are reported on the CAZy website, e.g., at http://afmb.cnrs-mrs.fr/CAZY/index.html.

5. The CAZy website reports a total of twelve GH74 catalytic domain entries. Of these, only 3 have been isolated and characterized. The remaining 9 entries included open reading frames or putative genes from genomics publications.

6. Two such entries are from thermophiles, including *Caldicellulosiruptor sp.* Tok7B1 and *Thermota maritima*; however, these genes have not been cloned or expressed, which has prevented the enzymes from being tested for thermal tolerance.

7. Exhibit A shows that no GH74 catalytic domain is closer than 53% identity with respect to SEQ ID NO. 3. These results show that SEQ ID NO. 3 represents a unique composition of matter.

8. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like are punishable by fine or imprisonment, or both, under 18 U.S.C. § 1001, and

that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Date: 6-17-03

By: Michael E. Himmel

Michael E. Himmel, PhD.

Family 74 BLAST with Acell F74

Putative secreted cellulase [SCO6545] [Streptomyces coelicolor]
Score = 841 bits (2172), Expect = 0.0
Identities = 402/754 (53%),

Glycosyl hydrolase 5 (Fragment) [Caldicellulosiruptor sp. Tok7B.1]
Score = 722 bits (1863), Expect = 0.0
Identities = 361/749 (48%),

Probably secreted sialidase, several ASP-boxes and dockerin domain [CAC0919] [Clostridium acetobutylicum]
Score = 718 bits (1854), Expect = 0.0
Identities = 359/753 (47%),

Endoglucanase C [EGLC] [Aspergillus niger]
Score = 630 bits (1624), Expect = e-179
Identities = 329/743 (44%)

CEL6 protein precursor [CEL6] [Agaricus bisporus (Common mushroom)]
Score = 618 bits (1593), Expect = e-175
Identities = 317/740 (42%

Avicelase III [AVIIII] [Aspergillus aculeatus]
Score = 597 bits (1538), Expect = e-169
Identities = 321/737 (43%)

Cellulase [CELA] [Xanthomonas axonopodis (pv. citri)]
Score = 377 bits (969), Expect = e-103
Identities = 247/748 (33%)

Cellulase [XCC1752] [Xanthomonas campestris (pv. campestris)]
Score = 369 bits (948), Expect = e-101
Identities = 243/747 (32%)

Endoglucanase, putative [TM0305] [Thermotoga maritima]
Score = 366 bits (940), Expect = e-100
Identities = 240/747 (32%)

Oligoxyloglucan reducing end-specific cellobiohydrolase [Geotrichumsp. M128]

Score = 357 bits (917), Expect = 3e-97
Identities = 255/806 (31%

Exhibit A